

UNITED STATES DEPARTMENT OF AGRICULTURE
AGRICULTURAL RESEARCH SERVICE

in cooperation with

STATE AGRICULTURAL EXPERIMENT STATIONS

Results from the

UNIFORM OATS WINTER HARDINESS NURSERY

2009-2010

Compiled by

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This is a joint progress report of an investigation underway in the State Agricultural Experiment Stations and the Agricultural Research Service of the U. S. Department of Agriculture. It contains preliminary data which have not been sufficiently confirmed to justify general release; interpretations may be modified with additional experimentation. Confirmed results will be published through established channels. The report is primarily a tool for cooperators, their staff and those with special interest in agricultural research program development.

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COOPERATING AGRICULTURAL EXPERIMENT STATIONS AND PERSONNEL

Country	State	AES Location	Personnel
USA	AR	Fayetteville	J. Kelly
USA	IL	Lawrenceville	L. Phillippe
USA	SC	Clemson	B. Edge
USA	NC	Laurel Springs/Waynesville	D. Marshall/M. Fountain
USA	TN	Knoxville	D. West
USA	LA	Baton Rouge	S. Harrison
Turkey	K.Maras	KSU Field Crops Department	T. Dokuyucu
Lithuania	Akademija	Lithuanian Univ. of Agriculture	A. Sliesaravicius
Poland	Blonie	Plant Breeding and Acclimatization Ins.	B. Lapinski
Czech Republic	Kromeriz	Agricultural Research Institute	L. Nedomova
Hungary	Martonvasar	Agric. Res. Inst. of Hungary Academy	O. Veisz
UK	Aberystwyth	IGER Plas Gogerddan	S. Cowan
Austria	Edelhof	Saatzucht Edelhof	S. Berger

DIGEST

NUMBER OF TESTS:	13 tests (6 US States, 7 foreign countries)	
NUMBER OF ENTRIES:	13	
EXPERIMENTAL DESIGN:	Single-row, 5-foot plot Two replications Randomized complete block	
DATA RECORDED:	Percent winter survival	
DATA NOT USED IN ANALYSIS:	Laurel Springs, NC	100% Survival
	Baton Rouge, LA	100% Survival
	Fayetteville, AR	100% Survival
	Akademija, Lithuania	0% Survival
	Knoxville, TN	No Data
	Clemson, SC	Data not received
	Lawrenceville, IL	Data not received
	Martonvasar, Hungary	Data not received
	Afsin, Turkey	Not planted

COMMENTS:

- Analysis of markers associated with winter hardiness was implemented beginning with the 2008-2009 nursery.
- All new oat lines will be evaluated with Simple sequence repeats (SSRs) associated with winter hardiness traits and continue to be added to the report.

US STATE/COUNTRY	LOCATION	COOPERATORS' COMMENTS
Poland	Radzików	The winter was with thick and long lasting snow cover. Snow mould occurred abundantly in rye and triticale trials; oat plots were highly or completely resistant.
Czech Republic	Kromeriz	Low germination of the standards.
Lithuania	Akademija	This winter was very cold. The temperature was 25-29 below zero for a long time.
UK	Aberystwyth	There was a marked difference in the 2 reps with rep 1 having better establishment and survival. There was no germination for plot 16 and plot 20.
Austria	Edelhof	Date of sowing November 2, 2009 due to very moist conditions. Precipitation more than long term average especially during sowing season. Closed snow layer from beginning of January till end of March. During this time, very low temperatures for a few weeks.

Table 1. Entries in the 2009-2010 Uniform Oat Winter Hardiness Nursery.

Entry No	Entry name	Pedigree	Yrs in Nursery	Contributors	
1	Fulgum (ck)	CI 708	72		
2	Norline (ck)	CI 6903	49		
3	Winter Turf (ck)	CI 3296	69		
4	Wintok (ck)	CI 3424	69		
5	NC03-2567v	H441/NC88-1652/NC88-1652	2	Murphy	NC
6	NC05-5460y	NC95-6073//TAM397/NC93-5856	2	Murphy	NC
7	NC07-3801lb	SS76-40/TAM397	1	Murphy	NC
8	NC07-3843y	Rodgers/SS76-40	1	Murphy	NC
9	Win/Nor-1	Wintok x Norline	7	Livingston, Murphy	NC
10	Win/Nor-10	Wintok x Norline	8	Livingston, Murphy	NC
11	Win/Nor-10b	Selection from Win/Nor-10	6	Livingston, Murphy	NC
12	ACS833	833 - released variety	2	Harrison	LA
13	LA604	LA604 - released variety	2	Harrison	LA
14	LA99017	TX96M1385/LA604	1	Harrison	LA
15	LA02065SBSBSBSB-88	LA604//ACS833/TAMO397	1	Harrison	LA
16	PR-4H8	(Wintok x Avena macrostachya B6) x Leggett95-43Cn	1	Lapinski	Poland

Top Ten Ranked Survival Entries

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Top 10 ranked survival entries for 2009-2010

Rank	Ent No.	Entry	Pedigree	% Survival (across locations)
1	16	PR-4H8	(Wintok x Avena macrostachya B6) x Leggett95-43Cn	97
2	10	Win/Nor-10	Wintok x Norline	94
3	8	NC07-3843y	Rodgers/SS76-40	93
4	7	NC07-3801lb	SS76-40/TAM397	91
5	14	LA99017	TX96M1385/LA604	91
6	6	NC05-5460y	NC95-6073//TAM397/NC93-5856	91
7	11	Win/Nor-10b	Selection from Win/Nor-10	90
8	12	ACS833	833 - released variety	90
9	9	Win/Nor-1	Wintok x Norline	90
10	5	NC03-2567v	H441/NC88-1652//NC88-1652	86
LSD (0.05)				9

Table 2a. Winter Oat Survival (%) at Various Stations (sorted by entry number)

Ent. No.	Entry Name	Ranked Means	Means across loc	Radzikow Poland	Kromeriz Czech Republic	Edelhof Austria	Aberystwyth UK
1	Fulgum (ck)	16	36	45	22	0	77
2	Norline (ck)	13	81	94	75	60	95
3	Winter Turf (ck)	15	48	81	46	0	66
4	Wintok (ck)	14	49	83	17	0	94
5	NC03-2567v	10	86	71	86	95	93
6	NC05-5460y	6	91	96	81	95	92
7	NC07-3801lb	4	91	87	93	90	96
8	NC07-3843y	3	93	92	85	100	94
9	Win/Nor-1	9	90	94	91	90	83
10	Win/Nor-10	2	94	90	99	90	99
11	Win/Nor-10b	7	90	98	83	80	100
12	ACS833	8	90	100	85	90	86
13	LA604	11	84	75	95	75	100
14	LA99017	5	91	94	97	80	94
15	LA02065SBSBSBSB-88	12	83	86	79	70	96
16	PR-4H8	1	97	100	95	95	100
Average			81	87	77	69	92
LSD (0.05)			9	13	24	23	20
CV(%)			5	7	15	16	11

Table 2b. Winter Oat Survival (%) at Various Stations (sorted by rank)

Ent. No.	Entry Name	Ranked Means	Means across loc	Radzikow Poland	Kromeriz Czech Republic	Edelhof Austria	Aberystwyth UK
16	PR-4H8	1	97	100	95	95	100
10	Win/Nor-10	2	94	90	99	90	99
8	NC07-3843y	3	93	92	85	100	94
7	NC07-3801lb	4	91	87	93	90	96
14	LA99017	5	91	94	97	80	94
6	NC05-5460y	6	91	96	81	95	92
11	Win/Nor-10b	7	90	98	83	80	100
12	ACS833	8	90	100	85	90	86
9	Win/Nor-1	9	90	94	91	90	83
5	NC03-2567v	10	86	71	86	95	93
13	LA604	11	84	75	95	75	100
15	LA02065SBSBSBSB-88	12	83	86	79	70	96
2	Norline (ck)	13	81	94	75	60	95
4	Wintok (ck)	14	49	83	17	0	94
3	Winter Turf (ck)	15	48	81	46	0	66
1	Fulgum (ck)	16	36	45	22	0	77
Average			81	87	77	69	92
LSD (0.05)			9	13	24	23	20
CV(%)			5	7	15	16	11

**Table 3. Uniform Oats Winter Hardiness Nursery
Under Controlled Environment Freeze Test**

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Entry #	Entry Name	Survival Rating ¹	% Survival ²
1	Fulgum (ck)	0.6	29
2	Norline (ck)	2.7	89
3	Winter Turf (ck)	1.4	63
4	Wintok (ck)	2.9	96
5	NC03-2567v	0.8	33
6	NC05-5460y	1.3	51
7	NC07-3801lb	2.0	78
8	NC07-3843y	1.0	45
9	Win/Nor-1	1.9	78
10	Win/Nor-10	2.4	84
11	Win/Nor-10b	2.9	96
12	ACS833	2.0	79
13	LA604	2.0	77
14	LA99017	1.6	62
15	LA02065SBSBSB-88	1.4	58
16	PR-4H8	2.3	78
Average		1.8	68
LSD (5%)		0.41	18.1
CV		10.6	12.4

Parameters:

- 2 reps/10 plants per rep planted in cone-tainers (Livingston et al. 2005, Crop Science, 45:1545-1558)
- 5 weeks at 13°C; 12 hours light/dark period; 400µmole light intensity
- 3 weeks at 3°C; 12 hours light/dark period; 350µmole light intensity
- 3 days @ -3°C in the dark (subzero acclimation)
- Frozen @ 1°C/hour to -12°C for 3 hours
- Thawed @ 2°C/hour to 3°C
- Plants were watered once with 0.001% (v/v) Vitavax fungicide solution
- Plants were allowed to recover for 3 weeks in the greenhouse
- Plants were rated for regrow after 21 days by visually assessing leaves and roots.

¹Rating:

- 0 = Completely dead
- 1 = 1 survived (green) shoot or 1 primary root
- 2 = 1 or 2 survived (green) shoots or 1 survived shoot and 1 or 2 primary roots
- 3 = 1 or 2 survived shoots with developed roots (primary and secondary roots)
- 4 = 95% survived shoots with well developed roots
- 5 = 100% survived with very little or no sign of freeze damage; same as unfrozen plants

²Survival (%):

- 50% of plants with rating of 1plus all plants rated >2 divided by total number of plants frozen multiplied by 100

Marker-Assisted Selection (MAS)

Winter hardiness is related to multiple quantitative traits, including winter field survival, crown freezing tolerance, vernalization response, and photoperiod. Crown freezing tolerance (CFT) is measured in controlled freeze tests and is an important component to winter hardiness. Photoperiod (PPD) and vernalization response (VRN) are frequently correlated with winter field survival and freezing tolerance because these traits contribute to delaying new growth until after the danger of freezing temperatures has passed. This avoidance response, affected by the combination of certain photoperiod and vernalization traits, can be useful for increasing winter field survival. The 7C-17 translocation is thought to contain a cluster of genes for increased tolerance to freezing temperatures and has been significantly correlated with winter field survival and crown freezing tolerance.

Genetic markers are fragments of DNA that are linked with known genes or traits. Associating markers with winter hardiness component traits (above) provides a valuable tool for oat research programs. Simple sequence repeats, known as SSRs or microsatellites, are a popular marker choice due to their relative low cost and ease of use. Oat SSR markers were previously evaluated for their association with winter hardiness component traits in a ‘Fulghum’ x ‘Norline’ population and in an oat association mapping population consisting of 25 spring sown, 36 fall sown, and two facultative oat lines. The markers associated with selected traits were chosen for testing with lines from the Uniform Oats Winter Hardiness Nursery, and the data are presented in Table 4.

If the DNA fragment associated with the phenotype of interest is present, this suggests that the line may have the winter hardiness trait. For example, the SSR marker HVM20 is associated with crown freeze tolerance and the 7C-17 translocation. The presence of the HVM20-142bp allele could translate to increased crown freeze tolerance and would be desirable in a marker-assisted selection program.

In the case of winter hardiness, a combination of traits is necessary, and marker selection at this stage is preliminary. Even though entries in the nursery may be winter hardy, they may not necessarily possess all the winter hardiness component markers. Further research will clarify which combination of traits, and therefore which SSR markers, are most informative for the development of a marker-assisted selection program.

Table 4. Markers Associated with Winter Hardiness Traits

Entry No.	Primer			AM1	AM2	AM102	AM270 S-1 ⁴	HVM20 ⁴	JAO4042 ²	JAO4234
	Traits ¹			CFT	CFT	WFS, CFT	WFS, CFT	CFT	VRN	VRN, CFT
	Allele Size (bp)			218	164	220	206	142	262	260
	Entry name	# of Significant Alleles Associate with Winter Hardiness	Field % Surv.							
1	Fulghum	2	36	yes	no	no	no	no	no	no
2	Norline	9	81	no	no	yes	yes	yes	yes	no
3	Winter Turf	7	48	yes	yes	yes	no	no	yes	no
4	Wintok	9	49	no	yes	yes	yes	yes	yes	no
5*	NC03-2567v	4	86	-	no	yes	yes	yes	yes	no
6*	NC05-5460y	5	91	-	no	yes	yes	yes	yes	no
7	NC07-3801lb	8	91	no	no	yes	yes	yes	yes	no
8	NC07-3843y	9	93	yes	yes	yes	yes	yes	yes	no
9*	Win/Nor-1	9	90	-	yes	yes	yes	yes	yes	yes
10*	Win/Nor-10	9	94	-	yes	yes	yes	yes	yes	yes
11*	Win/Nor-10b	9	90	-	yes	yes	yes	yes	yes	yes
12*	ACS833	8	90	-	yes	yes	yes	yes	yes	yes
13*	LA604	8	84	-	no	yes	yes	yes	yes	yes
14	LA99017	10	91	yes	no	yes	yes	yes	yes	no
15	LA02065SBSBSBSB-88	9	83	yes	yes	yes	yes	yes	yes	no
16	PR-4H8	10	97	no	yes	yes	yes	yes	yes	no

* Data from 2008-2009 Uniform Oat Winter Hardiness Nursery SSR analysis. (-) indicates markers that were not included in last year analysis.

Table 4 (cont.)

	Primer	JAO4636	VRN1 ³	Xncl5-3 ⁴	AME23	AME168 ⁴	AME178	AME184
	Traits ¹	CFT	VRN	CFT	MAT, PPD, VRN	CFT	CFT	WFS, MAT, PPD, VRN
	Allele Size (bp)	286	390	232	263	224	182	193
Entry No.	Entry name							
1	Fulghum	no	no	no	no	no	no	yes
2	Norline	yes	yes	yes	yes	yes	no	no
3	Winter Turf	no	no	no	yes	yes	no	yes
4	Wintok	yes	yes	yes	no	no	no	yes
5*	NC03-2567v	-	no	-	no	no	-	no
6*	NC05-5460y	-	no	-	no	yes	-	no
7	NC07-3801lb	no	no	yes	no	yes	yes	yes
8	NC07-3843y	no	no	yes	no	yes	no	yes
9*	Win/Nor-1	-	yes	-	yes	yes	-	no
10*	Win/Nor-10	-	yes	-	yes	yes	-	no
11*	Win/Nor-10b	-	yes	-	yes	yes	-	no
12*	ACS833	-	no	-	yes	yes	-	no
13*	LA604	-	no	-	yes	yes	-	no
14	LA99017	yes	no	yes	no	yes	yes	yes
15	LA02065SBSBSB-88	no	no	yes	no	yes	no	yes
16	PR-4H8	yes	yes	yes	no	yes	no	yes

* Data from 2008-2009 Uniform Oat Winter Hardiness Nursery SSR analysis. (-) indicates markers that were not included in last year analysis.

Footnotes for table 4.

Entries in the 2009-2010 Oat Winter Hardiness Nursery were evaluated with SSR and PCR markers associated with winter hardiness traits identified by Maloney et al. (submitted). Alleles shown are associated with positive effects on winter hardiness traits in the “Fulghum” x “Norline” mapping population and the oat association mapping population.

1. **WFS** = Winter field survival. Winter field survival was estimated after spring re-growth as the percent survival for the plots corrected for plot variation in germination or fall growth.
PPD = Photoperiod. Includes long and short photoperiod effects after 42 days of differing photoperiod treatments. Photoperiod, combined with vernalization and heading date, can mediate avoidance of freezing temperatures.
VRN = Vernalization. Vernalization response to differing temperature treatments during seed germination, as described in Wooten et al. (Crop Sci (2009) in press). Vernalization time, along with photoperiod and heading date, may result in avoidance of freezing temperatures.
CFT = Crown Freeze Tolerance. Crown freezing tolerance was evaluated by preparing and freezing crowns in a controlled environment chamber as described by Wooten et al. (2009). After three weeks of re-growth, recovery for each crown was visually measured on a scale of 0-10 (0=complete plant death, 10=no freezing damage).
2. JAO primer sequences may be requested from Joe Anderson at USDA-ARS, Purdue University.
3. VRN1 fragments are amplified by PooidVRN1 and Asintron-Rev in: Preston, J.C. and Kellogg, E.A. (2008), Plant Physiol. 146, no. 1: 265-276. VRN1 is involved in regulating response to vernalization in wheat and oats.
4. Marker is associated with presence or absence of the translocation 7C-17. Fulghum non-T7C-17, Norline T7C-17. Presence of the translocation is associated with increased crown freezing tolerance (Wooten et al. Crop Sci 47:1832-1840 (2007)).